

The Effect of Purified Catechins from Gambier on the Physico-Chemical Characteristics of Functional Solid Shampoo

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Gambier is a natural product that contains high amounts of catechin. Catechin has bioavailability as an antimicrobial such as fungus. Dandruff is a problem on the human scalp caused by fungus where this condition can be treated by using shampoo. The purpose of this study was to ascertain how the properties of solid shampoo were affected by the catechin extract from Gambier. This study's design comprised three replicates and five treatments in a fully randomized fashion. Catechin extract was added to the shampoo in amounts of 0, 1, 2, 3, and 4% of the total contents. Evaluations were conducted on the created shampoo's chemical, physical, and organoleptic properties. By the measurement of pH, percentage of solid content, foam height, texture, color, and scent revealed that the addition of gambier catechin extract could had a substantial influence ($\alpha = 5\%$) but not for dirt dispersal. Adding 4% of Gambier's catechin extract to the solid shampoo resulted in the optimal concentration. Further studies are needed to ascertain the specific role of the Gambier catechin extract in solid shampoo in order to fully appreciate the benefits of using it in shampoo products.

Keywords: Antimicrobial, dandruff, flavonoid, functional, health care.

INTRODUCTION

Uncaria gambier Roxb, a plant often grown in Indonesia, has the potential to be therapeutic. A common plant in West Sumatra, North Sumatra, South Sumatra, and Riau is the gambier. A plant item with significant economic significance for Indonesia is the gambier since many industries might used it as raw material such as pharmaceutical, textile, food and etc. (Hamda, 2014). Regarding 80% of the global gambier supply, Indonesia is the leading producer; 90% of Indonesia's gambier comes from West Sumatra (Hendri *et al.*, 2021).

The gambier plant, a shrub that resembles coffee and is a member of the Rubiace family, is a good source of antioxidants since it contains polyphenolic chemicals. Gambier's primary constituents are quercetin, catechin (catechin acid), and catechin tannic acid (catechin anhydride). Catechins are sweet-tasting but can become bitter when heated for an extended period of time or when heated with an alkaline solution known as catechin tannate. Catechins, catechin tannic acid, pyrocatechol, Gambier fluorescent red catechin, quercetin, fixed oil, and wax are among the chemical components found in gambier extract. Catechins

and tannins are the most widely used chemical components of Gambier (Anggraini and Syukri, 2023).

The catechin content in gambier is a characteristic that determines the quality level and quality of Gambier extract. It is because catechins are the main substituent of Gambier with considerable industrial demand compared to tannins. The higher the catechin content, the better the quality of gambier. Catechins, in their pure state, give a sweet, crystalline taste, white to yellowish, while tannins taste astringent, reddish brown to blackish. The main components in Gambier plants that act as antioxidants and antimicrobials are catechins or polyphenols (Saad *et al.*, 2020).

The public favors using natural materials because they are considered safer, practical, and economical and have fewer side effects than chemicals. Extracts of Gambier can be used for beauty and skin health due to its beneficial effect (Nandika *et al.*, 2019). Gambier's catechin extract is good for skin health as an anti-aging active ingredient that can maintain skin firmness, anti-acne active ingredients, and sunscreen. These bioactive compounds are optimally used on the skin by minimizing particles and allowing them to penetrate the skin pores well (Ismail *et al.*, 2021). Gambier's catechins' capacity

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to attach to other chemical compounds, especially proteins, makes them effective antibacterial agents. Catechins can bind to proteins on bacterial cell membranes to form complex compounds that interfere with the function and role of cell membranes and even cause leakage and cell death (syarifah *et al.*, 2019).

According to skin health effect and antimicrobials of catechins, it can be further developed as an ingredient in shampoo. Shampoo is generally made from a mixture of natural ingredients (plants) with chemicals in the form of surfactants and salt. In the manufacture of shampoo, there are several purposes, for example, to eliminate dandruff, blacken hair, strengthen hair roots, and other special purposes related to healthy head skin and hair. Some types of shampoo can cause allergic reactions in certain people. Like soap, shampoo is not recommended for infants and children because it contains chemicals that cause stinging in the eyes and irritation to the scalp of infants and children. For this reason, the essential components and additional herbal ingredients used in shampoo can be adjusted to the function of the shampoo and its intended use (D'souza *et al.*, 2019; Couteau *et al.*, 2015).

The shampoo examined in this study represents a product that addresses hair dandruff. The presence of scalp disorders such as sensitive, oily, and dandruff can interfere with average hair growth (Alessandrini and Piraccini, 2016). The problem of dandruff is one of the causes of a person's reduced self-confidence in activities (Galliano *et al.*, 2023). It is informed that more than 60% of the population in the world experience dandruff hair problems.

Usually, people in Indonesia use shampoo products in liquid form. However, this kind of shampoo might be wasteful as well as ineffective for traveling. An alternative that can be developed is to make shampoo products in the form of bars or solid shampoo. Solid shampoo is a shampoo that is available in solid form, just like solid soap. The composition is the same as shampoo in general, which contains surfactants, hair conditioning agents, and other additives such as preservatives, fragrances, colorants, and the addition of anti-dandruff ingredients to support exceptional performance in shampoo (Chiu *et al.*, 2015).

This research was conducted because there was no solid shampoo product with the addition of Gambier catechin extract. There has yet to be any specific data about solid shampoo with Gambier extract. As well as seeing the effect of the addition of gambier catechin extract with various concentrations to see the impact of the addition on the characteristics of solid shampoo and its activity as an antifungal for anti-dandruff function.

MATERIALS and METHODS

This study was carried out at the Department of Agricultural Products Technology, Faculty of Agricultural Technology,

Universitas Andalas, Padang; the laboratories were the Laboratory of Biochemistry of Agricultural Products and Food Nutrition, Laboratory of Microbiology and Biotechnology, Laboratory of Engineering Technology and Process of Agricultural Products. The study was carried out between September 2022 and October 2023.

Equipment: The equipments used for analysis were analytical scales (Kern ABJ 220, Germany), universal pH paper (merck), mixer (Ultra thurax T25, IKA), mold press (pneumatic system), test tubes (pyrex), stirring rods (pyrex), mortar (pyrex) and pestle (pyrex), beakers (pyrex), measuring cups (pyrex), spoons (pyrex), digital scales (CAS JP), and shampoo containers (40 x 60 cm x 15 cm, Lion star).

Materials: The ingredients used in making Gambier shampoo were cetyl alcohol, stearic acid, sodium lactate, sp incoquat behenyl tms-50, coco betaine, sodium gluconate, phenoxyethanol, panthenol vit. B, geranium, polyquaternium-7, tea tree oil, natrium cocoyl isethionate, natrium methyl cocoyl taurate, and catechin's Gambier extract. The concentration of catechin was approximately 90%.

Formulation of Gambier shampoo: Table 1 shows the formulation that was utilized to make Gambier shampoo after catechin extract concentration was added.

Table 1. Gambier shampoo formulation.

Ingredients	Addition of catechin for gambier's extract				
	(0%A)	(1%=B)	(2%=C)	(3%=D)	(4%=E)
Cetyl alcohol (g)	93.5	93.5	93.5	93.5	93.5
Stearic acid (g)	16.5	16.5	16.5	16.5	16.5
Behentrimonium Methosulfate (and) Cetyl alcohol (and) Butylene Glycol (g)	16.5	16.5	16.5	16.5	16.5
Sodium Lactate (g)	16.5	16.5	16.5	16.5	16.5
Coco betain (g)	44.0	44.0	44.0	44.0	44.0
Sodium gluconate (g)	5.5	5.5	5.5	5.5	5.5
Panthenol Vit B (g)	5.5	5.5	5.5	5.5	5.5
Phenoxyethanol (g)	2.8	2.8	2.8	2.8	2.8
Polyquaternium-7 (g)	16.5	16.5	16.5	16.5	16.5
Sodium cocoyl isethionate (g)	264.0	264.0	264.0	264.0	264.0
Sodium methyl cocoyl taurate (g)	49.5	49.5	49.5	49.5	49.5
Uncaria gambier extract (g)	0.0	5.5	11.0	16.5	22.0

Description: The amount of added gambier catechin extract used is based on the percentage of total ingredients used, which was 544.6g.

Shampoo production: First, prepare all the ingredients according to the formula. Separate the components based on 3 phases. Phase A consists of 93.5 g of cetyl alcohol, 16.5 g of stearic acid, 16.5 g of sodium lactate and catechins gambier extract, and 16.5 g of sp incoquat behenyl tms-50. Phase B consists of 44 g of cocobetaine, 5.5 g of sodium gluconate, 2.8 g of Phenoxyethanol, 5.5 g of panthenol vit. B, 2.8 g of geranium, 16.5 g of polyquaternium-7, and 2.8 g of tea tree



oil. Phase C consists of 264 g of sodium cocoyl isethionate and 49.5 g of Sodium Methyl Cocoyl Taurate.

Phase A was cooked in a double boiler at 50°C until it melted. In a single vessel, the phase B material was blended; in a separate container, the phase C materials were also homogenized. The phase B mixture was then mixed with the melted phase A. After that, phase C was added to the phase A and B combinations and homogenized to create a semi-solid dough. Subsequently, a press mold was used to shape the shampoo dough.

Observations raw material analysis

Catechin Analysis: Using the catechin external standard, high-performance liquid chromatography (HPLC) was used to examine the catechins. A 10 mL volumetric flask containing 100 µL of catechin from gambier extract was filled with 4 mL of a 50% (v/v) acetonitrile solution in water. The mixture was then ultrasonicated under room temperature for 1 hour to a homogeneous solution. The solution was diluted with pure water (distilled water) up to the mark, then filtered with 0.45 µm hydrophilic membrane. Dilution (if required) was carried out using 20% (v/v) acetonitrile solvent (Kumamoto *et al.*, 2000; Rini *et al.*, 2021).

Produced shampoo analyses

Moisture content of raw materials: The gravimetric method was used to calculate the moisture content. For thirty minutes, a temperature of 105°C was used to dry clean, empty aluminum cups. After that, the cup spent fifteen minutes cooling in a desiccator. An aluminum cup containing two grams of test material was filled with water and baked for three hours at 105 degrees Celsius. After cooling in a desiccator, the sample was weighed. Until a consistent weight was reached, drying was repeated (Thammawong *et al.*, 2019; Syukri *et al.*, 2013).

pH Analysis: The pH analysis process proceeded simply the following: After rinsing the electrodes in water devoid of minerals, pat dry with a gentle cloth. Once the pH meter displays a stable value, dip the electrode into the test sample. Note the number or scale reading that appears on the pH meter's display. When measuring pH, note the temperature and report the findings. After measuring, rinse the electrode once again with water devoid of minerals (Syukri *et al.*, 2023).

Dispersion of dirt: Each shampoo bar was divided into two drops, one for each milliliter of distilled water in a test tube. They added one drop of India ink. After capping the test tube, it was shook for ten minutes. According to Kumar and Mali (2010), there was none, light, medium, or thick ink in the foam.

Percentage of solid content: To calculate the percentage solids content, weigh approximately 4g of shampoo bar in an evaporation dish. The weight of the shampoo and dish were calculated. The shampoo's liquid portion was evaporated by heating it in a mantle. Ultimately, the percentage of solid material was determined by weighing the solid content in the

shampoo bar after it had completely dried (Kumar and Mali, 2010).

Foam Height Test: A scaled tube containing 2 g of shampoo was filled with 10 mL of distilled water, closed, and shaken for 20 seconds. We measured the height at which the foam developed. Additionally, foam height and stability were noted both during and five minutes after the shaking process (Quadeib *et al.*, 2017).

Physical Stability Test: The shampoo was placed in an environment that varied in temperature from 4 °C to 40 °C. This was done to assess the shampoo's physical stability. The shampoo was stored at different temperatures in order to conduct the physical stability test. Four weeks of storage were used, with occasional observations. The shampoo's look was the main subject of the observations (Meshram *et al.*, 2013).

Antimicrobial Activity Test (Anusha *et al.*, 2008; Dandekar *et al.*, 2022)

Media fabrications: Sabouraud Dextrose Agar (SDA) medium was used for rejuvenation and as a medium for testing antimicrobial activity on *Malassezia furfur* yeast.

Rejuvenation of Test Microbes: Rejuvenation of the test microbes was carried out by taking the initial microbial culture as much as one ose and then scratching on the surface of the inclined agar. For 48 hours, the *Malassezia furfur* yeast was cultured at room temperature (20–25°C).

Staining of test microbes: Staining of test microbes is a procedure for coloring test microbes using dyes that can highlight certain structures of the test microbes to be observed. Staining of *Malassezia furfur* yeast using Lactophenol Cotton Blue (LPCB) dye. Positive results were indicated by microscopic images of oval-shaped cells with one cell or spherical shape sprouting.

Preparation of Microbial Test Suspension: A few doses of the rejuvenated *Malassezia furfur* yeast were incubated for 48 hours before being placed in a tube with 5 mL of sterile 0.9% NaCl solution. The turbidity was then equalized with Mc Farland solution, which was equivalent to a concentration of 108 CFU/mL, to prepare the test microbial suspension. Following that, 1 mL of the solution was diluted by adding it to a test tube holding 9 mL of 0.9% NaCl (107 CFU/mL).

Preparation of Extract Concentration Solution: Preparation of extract concentration solutions by making solid shampoo into a suspension form by means of, solid shampoo that has been mashed using a pestle and sterile mortar, after the shampoo was smooth, 10 mL of sterile distilled water was added and homogenized. The shampoo extract was then put into a small Petri dish and labeled for each concentration.

Antimicrobial Activity Testing (Mayser *et al.*, 2003)

Inhibition Diameter Testing: Inhibition diameter testing was carried out using paper discs by the pour plate method. Sterile PDA media of as much as 3 mL was poured into each petri dish, then the results of the *Malassezia furfur* yeast suspension were taken as much as 0.1 mL using a micropipette and then inserted into a sterile petri dish that already contained PDA



media and leveled using trials slowly. The agar medium was allowed to solidify. After the media was solid, a well was made using a micropipette, but not to the bottom of the media. Next, paper discs were placed in the wells that had been made on PDA media. In different sterile Petri dishes, as much as 0.1 mL of the test extract with a concentration of 0%, 1%, 2%, 3%, and 4% was dripped into a sterile disc and allowed to dry after the sterile disc was dry. It is placed on solid agar media that already contains test microbes. Petri dishes were tightly wrapped and incubated. The yeast was incubated at 37°C room temperature for 24 hours. Antimicrobial activity was observed by measuring the diameter of the clear zone around the disk using a caliper.

Minimum Inhibitory Concentration Testing: The Minimum Inhibitory Concentration test was carried out if the inhibitory diameter test showed an inhibition zone that can be seen and measured in diameter. The extract concentrations used for *Malassezia furfur* were 0%; 1%; 2%; 3% and 4%. The Minimum Inhibitory Concentration Test was carried out by pipetting the concentration of the extract and the test microbial suspension as much as 0.1 mL into a sterile Petri dish containing agar media, then leveled using trials. The media was incubated at 37°C for 24 hours. The observation of the Minimum Inhibitory Concentration Test was determined on the lowest extract concentration cup that was not overgrown with microbes.

Organoleptic Test (Rini et al., 2021): The organoleptic test conducted in this study was hedonic (favorability test) by 25 semi-trained panelists. The organoleptic test carried out was an acceptance test, where each panelist was required to express a response about the product presented. In this test, panelists were asked to express their responses to shape, color, and aroma. The hedonic scale used is to use 5 numerical scales, namely very like (5), like (4), usual (3), dislike (2), and very dislike (1).

RESULTS

Raw Material Analysis: Analysis of raw materials used in this study was the analysis of moisture content and catechin content in Gambier leaves. The analysis results can be seen in Table 2.

Tabel 2. Raw Material Analysis

Parameter Analysis	Gambier leaf catechins \pm SD
Catechin Content (%)	86.81 \pm 1.64
Water Content	15.48 \pm 1.47

The analysis of raw materials revealed a moisture content of 15.48% and a catechin content of 86.81%. The results of the raw materials analysis were similar to the existing literature (Anggraini et al., 2011). This finding was probably because, during the purification process of gambier catechin extract, there were some shortcomings, such as drying only at room

temperature so that the resulting water content was still relatively high.

The HPLC testing of catechin extracts to determine catechin levels resulting from the purification of catechin from gambier leaves differs from the purification of catechin from whole Gambier. Furthermore, the method used will also affect the levels of catechins produced. Water content testing was done to determine the effect of the water content of catechin raw materials on the shampoo produced.

Product Appearance: Figure 1 shows the appearance of the resulting shampoo product. An authentic color change effect was found due to adding gambier extract catechins to the resulting shampoo. Therefore, adding gambier extract catechin might affect the chemical, physical, and even organoleptic characteristics of the product produced. Therefore, further analyses need to be conducted.



Figure 1. Shampoo bar with addition of catechin from gambier

Chemical Observation of Catechin Solid Shampoo

pH Analysis: A common method to assess changes in a product's acidity and basicity is to analyze its pH value. The findings of the variance analysis indicate that the pH value of the solid shampoo generated is significantly affected, at the 5% level, by the addition of catechins during the manufacturing process. Table 3 displays the analysis's findings.

Table 3. Average pH Test Value of Solid Shampoo.

Treatment	Mean pH \pm SD
(E) gambier catechin extract addition 1%	5.71 \pm 0.01a
(D) gambier catechin extract addition 2%	5.72 \pm 0.02a
(C) gambier catechin extract addition 3%	5.72 \pm 0.01a
(D) gambier catechin extract addition 4%	5.73 \pm 0.01a
(E) gambier catechin extract addition 0%	5.80 \pm 0.01b
KK = 0.04%	

Table 3. In this research, the average pH value varied between 5.71 and 5.80. With a score of 5.80, treatment A (addition of 0% Gambier Catechin Extract) produced the highest average value. Treatment E (4% addition of catechin Gambier extract) yielded the lowest average value, 5.71. The pH that was obtained will drop more when gambier catechin extract was added.



According to the analysis's findings, treatment A had the highest pH and treatment E the lowest. The pH decreases with increasing gambier catechin extract administration. In the soap formula, the stearic acid concentration can affect the pH. The acid has a low pH level, which ranges from 6.0 - 7.5 at 20 g / 25 ° C, which causes the pH of solid shampoo to be classified as low. The addition of catechin extract causes the shampoo's pH to decrease because catechins generally have a reasonably low pH or are acidic. Low pH usually ranges from 3 to 5. Sodium lactate can be used to adjust the pH of shampoo products. It helps maintain a pH that is suitable for the human scalp. Stearic acid can also be used to adjust the pH of shampoo products. Proper pH regulation is important to maintain the stability of the product and ensure that the shampoo matches the appropriate pH of the human scalp. Sodium gluconate can be used to regulate and maintain the pH of shampoo products. This component is important because the pH corresponding to the human scalp is slightly acidic, about 4.5 to 5.5. Proper pH regulation helps maintain scalp balance and avoid potential irritation due to drastic pH changes (Gavazzoni *et al.*, 2014).

Dirt Dispersion Analysis: Dirt dispersal analysis measured how much the shampoo can remove or reduce dirt, oil, dust, and other particles from the hair and scalp. Table 4 displays the findings of the dirt dispersal analysis.

Table 4. Dirt Dispersion Analysis Results

Treatment	Amount of ink in foam
(E) gambier catechin extract addition 0%	None
(D) gambier catechin extract addition 1%	None
(C) gambier catechin extract addition 2%	None
(D) gambier catechin extract addition 3%	None
(E) gambier catechin extract addition 4%	None
KK = 0.04%	

In Table 4, it can be seen that each solid shampoo product produced does not leave ink in the foam, which means that the ability of solid shampoo to remove or reduce dirt is good. This result is because, in the formulation of making solid shampoo, there are ingredients that function as surfactants that function as oil and dirt insertion materials such as coco betaine, Sodium cocoyl isethionate, and Sodium methyl cocoyl taurate. Surfactants can bind oil and dirt from the hair and scalp. Because they have a hydrophobic (oil-attracting) part and a hydrophilic (water-attracting) part, surfactants help "break up" the oil and dirt from the hair and scalp so that it can be mixed in water (Rhein *et al.*, 2007).

Solid Content Percentage Analysis: To figure out the amount of solid component was in the solid shampoo, a solid content percentage study was done. The inclusion of catechins in the production of solid shampoo has a substantial impact on the value of the analysis of the percentage of solid content

produced, at the 5% level, according to the variance analysis results. Table 5 displays the analysis's findings.

Table 5. Solid Content Percentage Analysis

Treatment	Mean solid content \pm SD
(E) gambier catechin extract addition 0%	87.43 \pm 1.17a
(D) gambier catechin extract addition 1%	89.44 \pm 1.07b
(C) gambier catechin extract addition 2%	90.71 \pm 1.23bc
(D) gambier catechin extract addition 3%	91.63 \pm 0.72c
(E) gambier catechin extract addition 4%	92.15 \pm 0.61c
KK = 0.04%	

Table 5 illustrates that the range of percentages of solid content obtained in solid shampoo is 87.43% to 92.15%. Treatment E (4% Gambier Catechin Extract Addition) has the highest value of 92.15%, while treatment A (0% Gambier Catechin Extract Addition) has the lowest value of 87.43%. The percentage of solid content produced increases with the amount of Gambier catechin extract used.

The concentration of catechins in the product formulation will significantly impact the percentage of solid content. The higher the concentration of catechins, the more significant the contribution to the solid percentage in the shampoo. It might due to the solubility of catechin in water that catechin are not soluble in water. The type of shampoo will also affect the solid content percentage. For example, solid shampoos generally have a higher percentage of solid content than liquid shampoos because the formulation differs.

In addition to the concentration of catechins in the shampoo being a solid content ingredient, several other ingredients are solid content. Solid content ingredients in shampoo give structure and texture to the shampoo. Cetyl alcohol is a type of fatty alcohol used in hair and skin care products to provide a soft and creamy texture to the shampoo. Stearic acid is a saturated fatty acid that provides texture and stability to hair and skin care products. Behentrimonium Methosulfate (and) Cetyl Alcohol (and) Butylene Glycol. This combination is a softening agent usually used in shampoos to provide conditioning properties to the hair (Marsh *et al.*, 2017). Sodium lactate is the sodium salt of lactic acid and can be used as a moisture-balancing agent in hair care products. Panthenol (Vitamin B5) is an ingredient used to hydrate and treat hair. Polyquaternium-7 is an ingredient used as a conditioning agent in hair care products, such as shampoos. Sodium cocoyl isethionate is an ingredient used as a cleansing agent in shampoos that also helps to produce lather. Moreover, sodium Methyl Cocoyl Taurate is also a cleansing agent in shampoos and helps to produce lather.

Foam Height Analysis: Foam height analysis in shampoo measures and assesses the amount and quality of foam produced during use. It is a method used to evaluate how much a shampoo produces adequate foam during hair-washing. The variance analysis results show that adding



catechins to the manufacture of solid shampoo has a significant effect at the 5% level on the foam height value of the solid shampoo produced. The results of the foam height analysis can be seen in Table 6.

Tabel 6. Foam height (cm) analysis of produced shampoo.

Treatment	Mean foam height \pm SD
(E) gambier catechin extract addition 4%	4.93 \pm 0.21a
(D) gambier catechin extract addition 3%	5.27 \pm 0.12b
(C) gambier catechin extract addition 2%	5.50 \pm 0.20bc
(B) gambier catechin extract addition 1%	5.60 \pm 0.10cd
(A) gambier catechin extract addition 0%	5.83 \pm 0.15d
CV = 0.59%	

In Table 6, it can be seen that the foam height produced by solid shampoo ranges from 4.93% - 5.83%. The highest average value is in treatment E (Addition of Gambier Catechin Extract 4%) with a value of 4.93%, and the highest value is in treatment A (Addition of Gambier Catechin Extract 0%) with a value of 5.83%. With the addition of Gambier catechin extract, the value of foam height in solid shampoo is getting lower.

Ingredients in shampoo that function as foam-forming agents are usually referred to as saponifying or surfactant agents (Rivai *et al.*, 2017). These surfactant agents produce foam, allowing the shampoo to clean the hair and scalp more effectively. Catechins have oxygen-binding properties so they can inhibit foam formation by surfactants. Catechins interacting with oxygen can reduce the efficiency of foam formation by surfactants. The interaction between catechins or other compounds with other ingredients in the shampoo formulation can also affect the foam height of solid shampoo. The ingredients that usually play a role in forming foam in shampoo are cleaning agents or surfactants. Two ingredients that have a significant role in forming foam in shampoo are Sodium cocoyl isethionate, a type of surfactant used in hair care products, including shampoo, which helps produce a soft lather. It is effective in cleaning the hair and scalp. Sodium methyl cocoyl taurate is a surfactant used in hair care products. It helps cleanse the hair and scalp while producing a lather.

Physical Observations of Solid Shampoo

Analysis of Physical Stability: Physical stability analysis was conducted to determine the stability of shampoo products after 4 weeks of storage at low temperature, cold temperature and high temperature. The results of the physical stability analysis can be seen in Table 7.

Physical stability analysis of solid shampoo in Table 7. There was a color change in the shampoo product. Low temperatures will generally not damage or alter catechins directly. Low temperatures usually slow down chemical reactions, including oxidation, that can damage compounds like catechins. In general, catechins do not undergo significant changes at cold or low temperatures during proper

storage. Cold temperatures usually do not damage catechins directly and can even help slow down oxidation reactions that can damage the compound. High storage temperatures can affect gambier catechins' stability, as exposure to high temperatures can trigger chemical reactions, including oxidation reactions, that can damage these compounds (Kim *et al.*, 2020). In addition to the catechins in solid shampoo products, storage errors such as poor packaging, humidity control, and light exposure control can also affect the stability of gambier catechins.

Table 7. Physical Stability Analysis of Solid Shampoo.

Tr.	Storage temperature		
	4°C	25°C	40°C
A	Stationary color	Stationary color	Yellowing color
B	Stationary color	Stationary color	The hue turns brown
C	Stationary color	Stationary color	The hue turns brown
D	Stationary color	Stationary color	The hue turns brown
E	Stationary color	Stationary color	The hue turns brown

Analysis of Antimicrobial Activity: Inhibition zone testing helps assess the shampoo's effectiveness in controlling *Malassezia* fungi's growth. If the shampoo can create a large zone of inhibition against these fungi, this can be considered a sign that the product can help prevent or reduce skin problems caused by *Malassezia* fungi. The variance results show that adding catechins to the manufacture of solid shampoo has a significant effect at the 5% level on the inhibition zone of the solid shampoo produced. The results of the antimicrobial activity analysis can be seen in Table 8 and Fig. 2.

Table 8. Antimicrobial activity ((cm²) of solid shampoo.

Treatment	Area of zone of microbial inhibition \pm SD
(A) Gambier Catechin Extract Addition 0%	0.00 \pm 0.00a
(B) Gambier Catechin Extract Addition 1%	0.87 \pm 0.12ab
(C) Gambier Catechin Extract Addition 2%	1.04 \pm 0.13ab
(D) Gambier Catechin Extract Addition 3%	1.43 \pm 0.15c
(E) Gambier Catechin Extract Addition r 4%	5.34 \pm 1.15d
KK = 6.07%	

Table 8 and Fig. 2 show that the inhibition zone area produced by solid shampoo in the antifungal activity test ranged increased with the increasing of Gambier catechin extract. The zone area was from 0 cm² to 5.34 cm². The most expansive inhibition zone area was found in treatment E (Addition of Gambier Catechin Extract 4%) with an area of 5.34 cm², and the smallest inhibition zone area was found in treatment A (Addition of Gambier Catechin Extract 0%) with an area of 0 cm². The more the addition of Gambier catechin extract, the wider the inhibition zone produced.



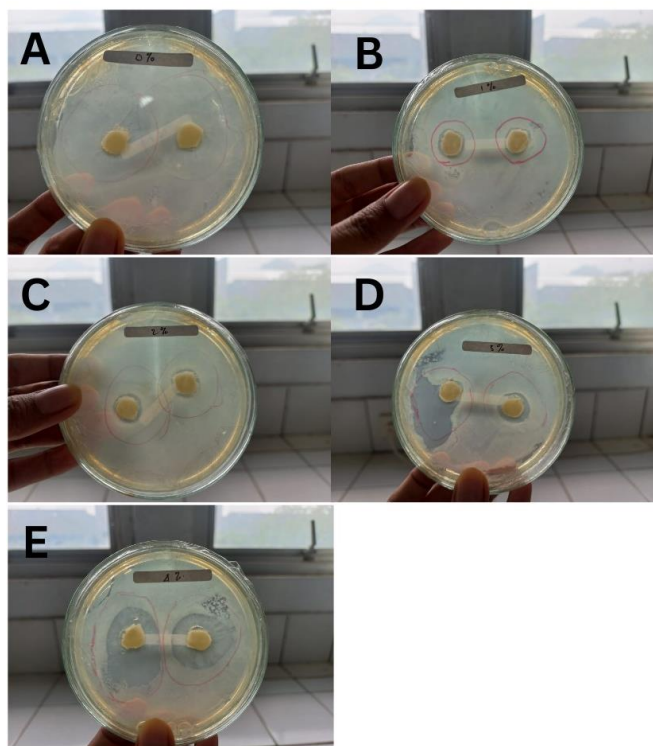


Figure 2. Inhibition zone of Gambier catechin extract against *Malassezia furfur*.

DISCUSSION

The use of catechins extracted from gambier sap can increase the bioactivity value of solid shampoo. Based on the results of physicochemical testing, a shampoo product was produced that meets the quality standards. The addition of catechins affects the physicochemical characteristics of the resulting product. Catechin is a substance in the form of a powder that has semi-polar properties. The nature of these catechins makes them not easy to dissolve in water, so the physical characteristics of these catechins can influence the chemical and physical properties of the product produced. In addition, products derived from catechins usually give extreme browning reactions during storage. This appearance may be due to the presence of tannin from gambier extract as a medium for catechins (Malrianti *et al.*, 2018). Therefore, making the product in bar or solid form provides better stability because the formation of brown color due to the presence of tannin will slow down.

Furthermore, using shampoo in solid form will make shampoo use more economical because the active substance content is in a concentrated concentration. This product was recommended to be used only a few times a week. From the perspective of anti-dandruff bioactivity, in this experiment, it was evident that with the presence of catechin, the inhibitory

power against dandruff precursor fungi is also greater. The ability of catechins to damage the activity of the *Malassezia* fungus is a positive value that has been hypothesized from the beginning of this research, and this research can prove it.

Catechins are antioxidant compounds that protect body and skin cells from damage caused by free radicals and oxidative stress. This antioxidant activity may contribute to the antimicrobial properties of catechins by helping to fight infection and fungal growth. Catechins have strong antifungal properties (Bernatoniene *et al.*, 2018; Michalak *et al.*, 2022; Messire *et al.*, 2023). This data means that catechins can interfere with the growth and development of fungi such as *Malassezia*. Catechins can also have anti-inflammatory effects that help relieve inflammation in the skin caused by fungal infections. This effect can help reduce symptoms such as itching, redness, and irritation. Catechins work by disrupting the integrity of the fungal cell membrane, inhibiting cell growth, and causing cell death (Jadimurthy *et al.*, 2023). This activity may help with infections caused by *Malassezia*.

The limitation of this research is that the analysis results were only carried out on an in vitro scale. However, this data can be a reference for developing prototype products that can be tested in the community. The resulting product can generally be developed towards the commercialization stage, where the shampoo product uses natural ingredients with anti-dandruff functionalities. To be marketed and beneficial to society, this product should undergo in vivo testing and practical applications.

Conclusion: The results showed that the addition of gambier catechin extract affected the pH, percentage of solid content, foam height, antimicrobial activity, texture, color, and aroma of produced shampoo. However, it did not significantly affect the solid shampoo's dirt dispersion and physical stability with the addition of Gambier catechin extract. Moreover, it was selected as the optimum treatment was the treatment E with the addition of 4% Gambier. Further research on the bioactivity of dandruff shampoo, such as in vivo analysis, needs to be considered. Moreover, this product can be tested further by receiving input from consumers regarding direct application in the community because the condition of the dandruff scalp varies from person to person.

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